

What is Claimed:

1. A method of identifying an environmental stimulus or a gene that alters the lifespan of an organism, said method comprising:
 - 5 providing a control cell culture and one or more test cultures, wherein the one or more test cell cultures but not the control cell culture comprise either (i) mother yeast cells that possess a genotype modification of either a non-essential gene or an essential gene, in which case the genotype modification is non-lethal, (ii) mother yeast cells that are exposed to an environmental stimulus other than a pro-oxidant, or
 - 10 (iii) mother yeast cells that possess a genotype modification of either a non-essential gene or an essential gene, in which case the genotype modification is non-lethal, and are exposed to an environmental stimulus other than a pro-oxidant;
 - culturing the control cell cultures and one or more test cell cultures under conditions whereby mother yeast cells can replicate and daughter yeast cells
 - 15 cannot; and
 - determining whether the mother yeast cells in the one or more test cell cultures exhibit a change in replicable lifespan when compared to the mother yeast cells in the control cell culture, wherein an increase in the replicable lifespan for mother yeast cells of a test cell culture indicates that the genotype modification, the
 - 20 environmental stimulus, or the combination thereof, enhances the replicable lifespan of the mother yeast cells in the test cell culture.
2. The method according to claim 1 wherein said growing is carried out in a growth medium that allows for mother cell replication but not
- 25 daughter cell replication.
3. The method according to claim 2 wherein the growth medium of the control cell culture and the one or more test cell cultures is free of galactose.
- 30 4. The method according to claim 2 wherein mother and daughter cells both possess two chimeric genes encoding a protein required for replication, one under control of a promoter responsive to growth medium conditions and the other under control of a promoter operable mother cells but not daughter cells.

5. The method according to claim 4 wherein the promoter responsive to growth medium conditions is a promoter responsive to presence of galactose.

5 6. The method according to claim 5 wherein the promoter responsive to galactose presence is a *GAL1*, *GAL7*, or *GAL10* promoter.

7. The method according to claim 4 wherein the promoter operable only in mother cells is an HO promoter.

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8. The method according to claim 4 wherein a native gene encoding the protein required for replication is disrupted to prevent expression of the protein therefrom.

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9. The method according to claim 4 wherein the protein required for replication is a cell cycle protein.

10. The method according to claim 9 wherein the cell cycle protein is selected from the group of *CDC2*, *CDC3*, *CDC4*, *CDC6*, *CDC7*, *CDC8*, *CDC9*,
20 *CDC10*, *CDC13*, *CDC16*, *CDC20*, *CDC23*, *CDC24*, *CDC26*, *CDC27*, *CDC28*,
CDC34, *CDC42*, and *CDC53*.

11. The method according to claim 1 wherein the one or more test cell cultures comprise mother cells that possess a genotype modification involving a
25 nonessential gene.

12. The method according to claim 11 wherein the genotype modification is selected from the group of a deletion mutant, an overexpression mutant, an addition mutant, or encoding a mutant protein.

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13. The method according to claim 1 wherein the one or more test cell cultures comprise cells that are exposed to an environmental stimulus other than a pro-oxidant.

14. The method according to claim 13 wherein the environmental stimulus is a mixture of natural or synthetic organic or inorganic products, plant or animal extracts, or tinctures, as well as combinations thereof.

5 15. The method according to claim 1 wherein the one or more test cell cultures comprise mother cells that possess a genotype modification involving a nonessential gene and are exposed to an environmental stimulus other than a pro-oxidant.

10 16. The method according to claim 1 wherein said determining comprises:

performing growth curve analyses on both the control cell culture and the one or more test cell cultures, and

15 assessing whether a difference exists between the growth curves of the control cell culture and the one or more test cell cultures.

17. The method according to claim 16 wherein said growing is carried out in a liquid growth medium.

20 18. The method according to claim 17 wherein said performing growth curve analyses is carried out by measuring optical density of the liquid growth medium containing the cells.

25 19. The method according to claim 1 wherein said determining comprises:

assessing colony size of colonies present in the control cell culture and colonies present in the one or more test cell culture, wherein colony size is equal to the replicable lifespan of the mother cell.

30 20. The method according to claim 19 wherein said growing is carried out on a solid growth medium.

21. The method according to claim 20 wherein said assessing is done manually.

22. The method according to claim 20 wherein said assessing is carried out by analyzing optical images.

23. The method according to claim 22 wherein said analyzing
5 optical images comprises:

capturing an image of colonies present in the control cell culture and an image of each of the one or more test cell cultures; and

calculating the two-dimensional area or a morphometric property of colonies in each of the images, wherein the two-dimensional area or the
10 morphometric property of a colony equates to the replicable lifespan of the mother cell.

24. The method according to claim 15 wherein the genotype modification is replacement of a yeast gene that regulates lifespan with a human
15 homolog of the yeast gene.

25. The method according to claim 24 wherein the human homolog is *RAS*, *BAX*, *SIR2*, *WRN*, or *BS*.

26. The method according to claim 1 wherein the yeast strain is a
20 homozygous diploid host strain of yeast carrying two identical copies of the first and second chimeric genes but having a mutation in one copy of the non-essential gene.

27. The method according to claim 1 wherein the one or more test
25 cell cultures comprises greater than ten test cell cultures.

28. The method according to claim 1 wherein the one or more test cell cultures comprises greater than one-hundred test cell cultures.

29. A DNA construct comprising first and second chimeric genes that both encode substantially the same protein that is required for yeast replication, the first chimeric gene comprising a promoter responsive to growth medium conditions and the second chimeric gene comprising a promoter operable in mother cells but not daughter cells.
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30. The DNA construct according to claim 29 wherein the promoter responsive to growth medium conditions is a promoter responsive to presence of galactose.

5 31. The DNA construct according to claim 31 wherein the promoter responsive to galactose presence is a *GAL1*, *GAL7*, or *GAL10* promoter.

32. The DNA construct according to claim 29 wherein the promoter operable in mother cells but not daughter cells is an HO promoter.

10 33. The DNA construct according to claim 29 wherein the protein required for replication is a cell cycle protein.

34. The DNA construct according to claim 33 wherein the cell
15 cycle protein is selected from the group of *CDC2*, *CDC3*, *CDC4*, *CDC6*, *CDC7*, *CDC8*, *CDC9*, *CDC10*, *CDC13*, *CDC16*, *CDC20*, *CDC23*, *CDC24*, *CDC26*, *CDC27*, *CDC28*, *CDC34*, *CDC42*, and *CDC53*.

35. An expression vector comprising the DNA construct according
20 to claim 29.

36. The expression vector according to claim 35 wherein the expression vector is a plasmid.

25 37. A host cell comprising the DNA construct according to claim 29.

38. The host cell according to claim 37 wherein the DNA construct is present within an expression vector.

30 39. The host cell according to claim 38 wherein the host cell is a bacterium.

35 40. The host cell according to claim 39 wherein the bacterium is *Escherichia coli*.

41. The host cell according to claim 37 wherein the host cell is a yeast.

42. The host cell according to claim 41 wherein the yeast is an
5 asymmetrically-dividing yeast.

43. The host cell according to claim 41 wherein the yeast is selected from the group of *Candida*, *Saccharomyces*, and *Schizosaccharomyces*.

10 44. The host cell according to claim 41 wherein the yeast comprises a disrupted native gene, which but for the disruption would express the protein required for yeast replication.

45. The host cell according to claim 41 wherein the DNA construct
15 is integrated into the genome of the yeast.

46. The host cell according to claim 41 wherein the DNA construct is present within an expression vector.

20 47. A kit comprising:
a first container comprising a first growth medium and yeast cells in the growth medium, the yeast cells possessing two chimeric genes both encoding substantially the same protein that is required for replication, one chimeric gene comprising a promoter responsive to growth medium conditions and the other
25 chimeric gene comprising a promoter operable in mother cells but not daughter cells, wherein the first growth medium induces expression of the chimeric gene under control of the promoter responsive to growth medium conditions;

a second container that contains a growth medium that strongly represses expression of the one chimeric gene comprising the promoter responsive to
30 growth medium conditions; and

empty containers for growing yeast cells obtained from the first container in the growth medium of the second container.

48. The kit according to claim 47 further comprising:
instructions for growing the yeast cells in the growth medium of the
second container and determining the replicable lifespan of mother cells.

5 49. The kit according to claim 48 wherein the instructions are for
performing the method where the yeast cells are exposed to an environmental
stimulus other than a pro-oxidant.

50. The kit according to claim 47 wherein the promoter responsive
10 to growth medium conditions is a promoter responsive to presence of galactose.

51. The kit according to claim 50 wherein the promoter responsive
to galactose presence is a *GAL1*, *GAL7*, or *GAL10* promoter.

15 52. The kit according to claim 47 wherein the promoter operable in
mother cells but not daughter cells is an HO promoter.

53. The kit according to claim 47 wherein the protein required for
replication is a cell cycle protein.

20 54. The kit according to claim 53 wherein the cell cycle protein is
selected from the group of *CDC2*, *CDC3*, *CDC4*, *CDC6*, *CDC7*, *CDC8*, *CDC9*,
CDC10, *CDC13*, *CDC16*, *CDC20*, *CDC23*, *CDC24*, *CDC26*, *CDC27*, *CDC28*,
CDC34, *CDC42*, and *CDC53*.

25 55. The kit according to claim 47 wherein the yeast cells comprise
a gene that promotes shortened replicative lifespan.

56. The kit according to claim 55 wherein the gene that promotes
30 shortened replicative lifespan is selected from the group of *RAS*, *BAX*, *SIR2*, *WRN*, or
BS.

57. The kit according to claim 55 wherein the gene that promotes
shortened replicative lifespan is a human gene that replaces a homologous yeast gene.

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58. A method of identifying quantitative trait loci for aging-related genes in yeast, said method comprising:

providing first and second yeast strains having different replicative lifespans, each strain comprising two chimeric genes both encoding substantially the same protein that is required for replication, one chimeric gene comprising a promoter responsive to growth medium conditions and the other chimeric gene comprising a promoter operable in mother cells but not daughter cells;

mating the first and second yeast strains to produce diploid cells;

inducing meiosis in the diploid cells to produce spores;
isolating and germinating the spores to produce haploid offspring;

culturing the haploid offspring under conditions whereby mother cells can replicate and daughter cells cannot;

determining the replicable lifespan of haploid offspring; and
identifying one or more quantitative trait loci associated with the replicable lifespan of the haploid offspring.